

Sensitivity of Freshwater Mollusks to Hydrilla-targeting Herbicides



Jennifer M. Archambault and W. Gregory Cope
NC State University
Department of Applied Ecology

Acknowledgments

Mollusk Collection/Propagation

Chris Barnhart, Missouri State U.

Tom Fox

Jay Levine

Steve Hoyle

Justin Nawrocki

Cody Hale

North Carolina State Parks

Analysis/Laboratory/Field

Haywood Perry, SePRO

Angela White

SePRO

USACE

Funding

Gulf States Marine Fisheries

Commission/US FWS

Region 4

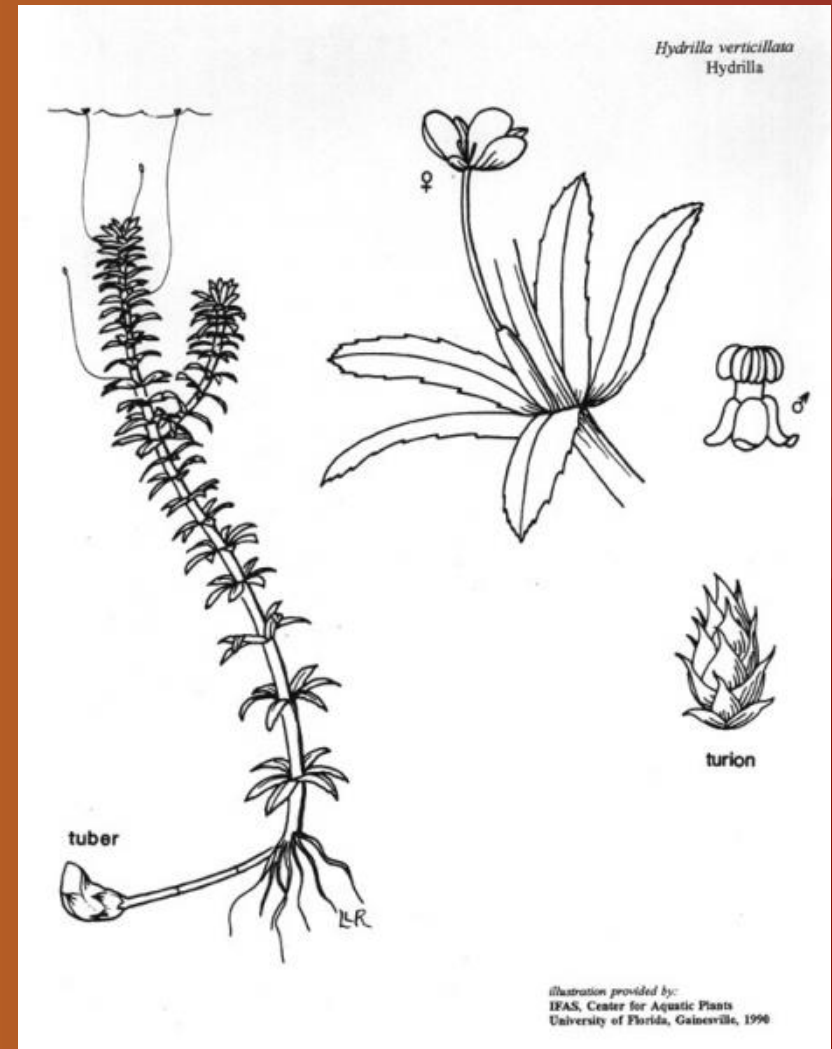
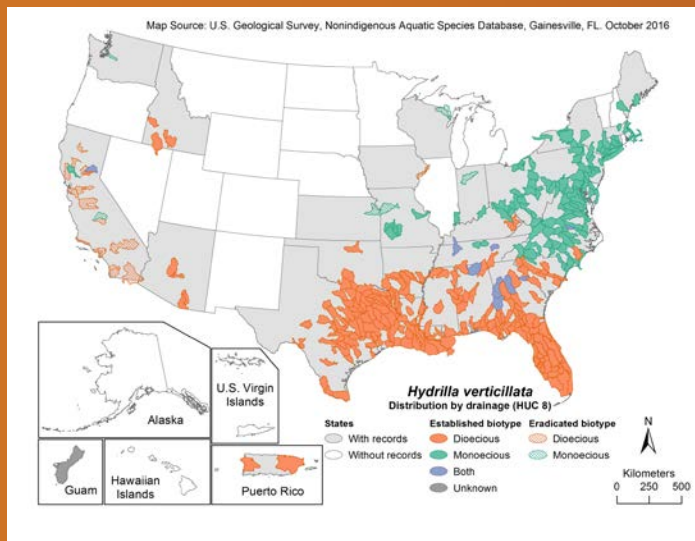


NC STATE UNIVERSITY



Hydrilla verticillata

- Hydrilla, water-thyme
- Monoecious biotype
 - Spreading by 1970s
 - Tubers, turions, fragments
- Expanding in range
 - Boats, angler equipment



Hydrilla verticillata

- Federal Noxious Weed in US
 - Monocultures
 - Alters water quality
 - Neurotoxic bacteria & AVM



Hydrilla verticillata

- **Federal Noxious Weed in US**

- Monocultures

- Alters water quality

- Neurotoxic bacteria & AVM

- **Control Strategies:**

- Mechanical removal

- Grass carp

- Herbicides

- Sonar (fluridone)

- Aquathol (endothall)



Herbicide Mode of Action

- **Sonar (fluridone)**
 - Chlorophyll / carotenoid pigment inhibitor
 - Prohibits plants from making food
- **Aquathol (endothall)**
 - Protein phosphatase inhibitor (interferes with respiration)



Freshwater Mollusks

- Extremely imperiled taxa
- Poorly studied
 - Knowledge far behind fishes, mammals, birds...
 - Still need basic science
- Sensitive to contaminants



FWGNA

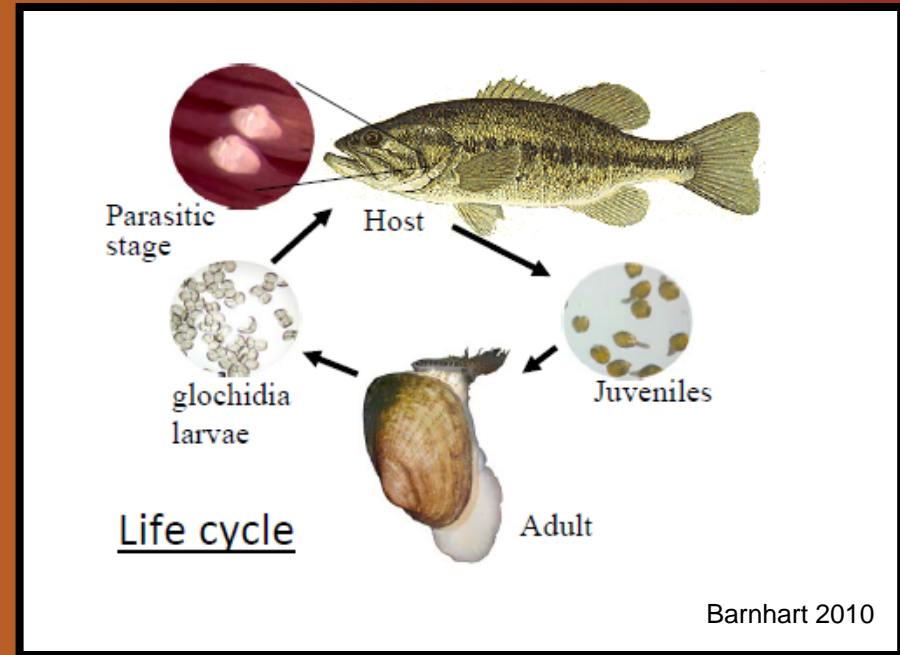


MC Barnhart



Unionid Mussels

- Found in streams, rivers, lakes, and ponds
- 673 species in Unionidae
~ half in N. America
- > 71% of species imperiled
- Water purifiers
- Unique life history –
obligate fish parasite



Gastropods (Snails)

- Found in streams, rivers, lakes, and ponds
- 703 species in US and Canada
- > 74% of species imperiled
- Highly susceptible to habitat loss and degradation

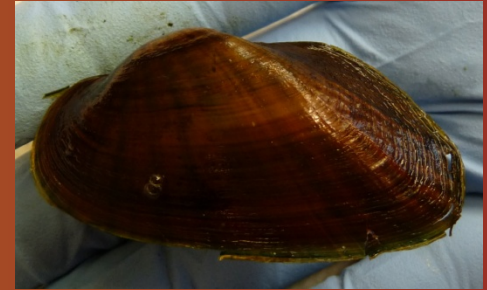


FWGNA



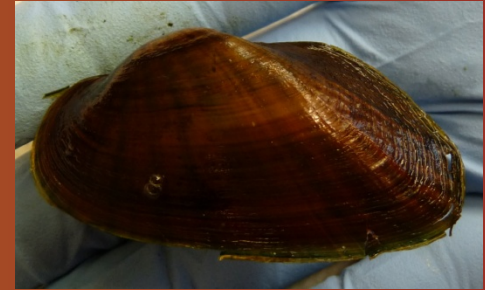
Lake Waccamaw

- 9,000 acre Carolina Bay lake
- Several endemic species
 - 3 fishes – W. silverside, W. killifish, W. darter
 - 2 mussels – W. fatmucket and W. spike
 - 2 snails – W. amnicola and W. siltsnail



Lake Waccamaw

- 9,000 acre Carolina Bay lake
- Several endemic species
 - 3 fishes – W. silverside, W. killifish, W. darter
 - 2 mussels – W. fatmucket and W. spike
 - 2 snails – W. amnicola and W. siltsnail
- Hydrilla recently introduced near boat ramp
 - ~ 600 acres infested

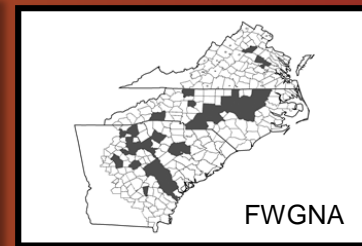


Eno River

- NC Piedmont stream, rocky, cobble substrates
- Panhandle pebblesnail present
- Imperiled (NCWAP species; G2)
- Rocky riffles with good flow
- Solid substrate
- Plentiful riffleweed (snail habitat)
- Hydrilla present



A. White



Research Needs & Objectives

Few data on invertebrates

No data for freshwater mollusks



Determine sensitivity of native mollusks to aquatic herbicide formulations

Sonar – Precision Release (PR) and Genesis (i.e., fluridone)

Aquathol – K (i.e., endothall)

Consider results in context of proposed treatment of *Hydrilla*

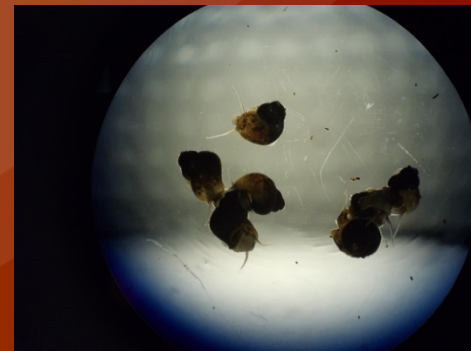
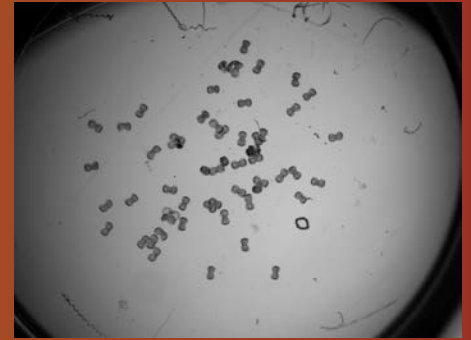
Acute Toxicity Experiments

- Fatmucket (*Lampsilis siliquoidea*)

- glochidia and juveniles

- Panhandle Pebblesnail (*Somatogyrus virginicus*)

- juveniles and adults



Acute Toxicity Experiments

- Fatmucket (*Lampsilis siliquoidea*)

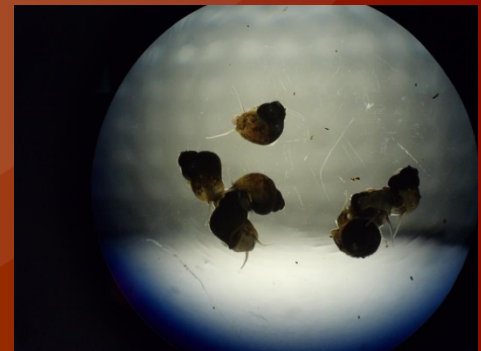
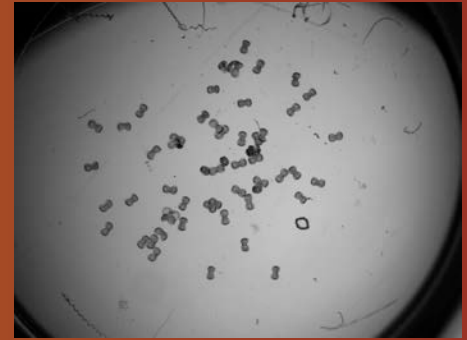
- glochidia and juveniles

- Panhandle Pebblesnail (*Somatogyrus virginicus*)

- juveniles and adults

- Follows ASTM (2006) Standard Guideline for Conducting Toxicity Tests with Mussels

- 24 h – glochidia, 96 h – juveniles and snails



Acute Toxicity Experiments

- Fatmucket (*Lampsilis siliquoidea*)

- glochidia and juveniles

- Panhandle Pebblesnail (*Somatogyrus virginicus*)

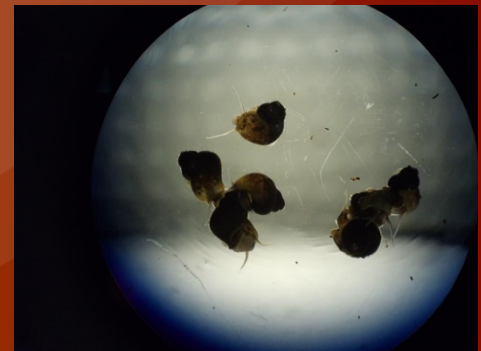
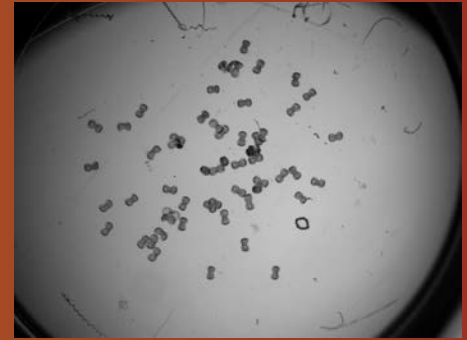
- juveniles and adults

- Follows ASTM (2006) Standard Guideline for Conducting Toxicity Tests with Mussels

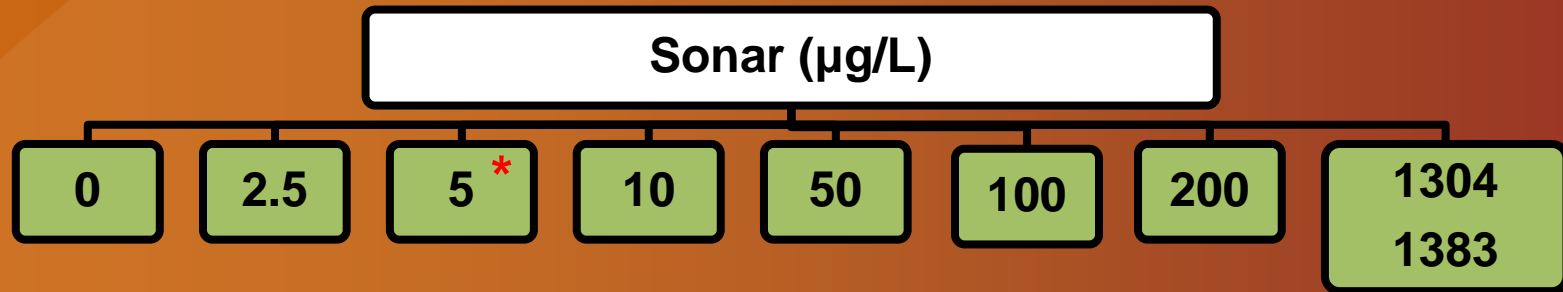
- 24 h – glochidia, 96 h – juveniles and snails

- Endpoint – survival

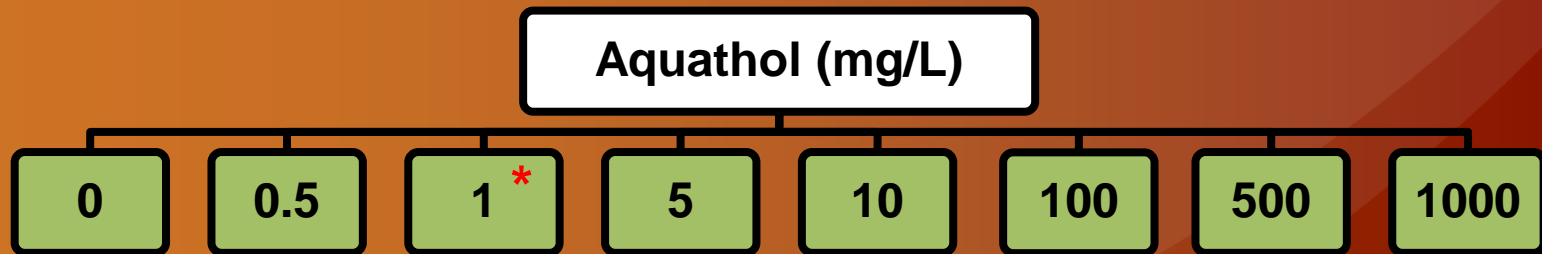
- Glochidia = shell closure response to NaCl
- Snails and juvenile mussels = movement in 5 min
- Median Lethal Concentration (LC50)



Acute Toxicity Experiments



Experiment with adult snails achieved maximum exposure concn. of 564 µg/L.



* Target application concn.

3 replicates/ treatment

28-d Chronic Experiment

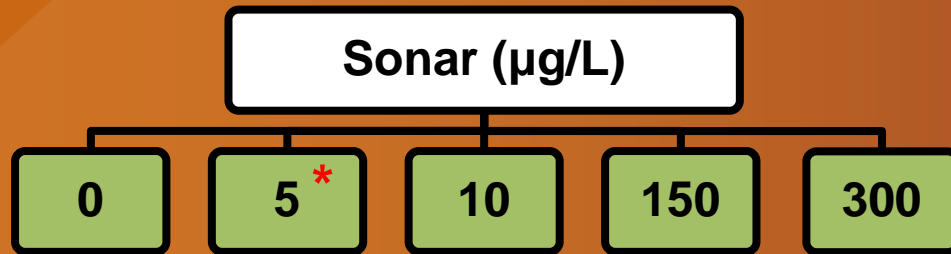
- Waccamaw fatmucket (*L. fullerkati*)
- Static test, aerated, 500 mL, renewed at 72 h



- Endpoints
 - Survival – 7, 14, 21, 28 d
 - Siphoning – # siphoning/exposed
 - Foot protrusion – # extended/exposed



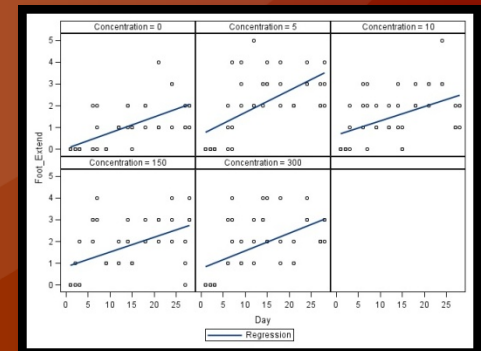
28-d Chronic Experiment



*** Target application concn.**

Statistical Analysis

- Viability / Survival:
 - LC50, Spearman-Kärber method (CETIS)
- Siphoning, Foot Protrusion, Snail Egg Hatching
 - Repeated measures ANOVA (PROC MIXED, SAS v. 9.3)
 - Dunnett's post-hoc test



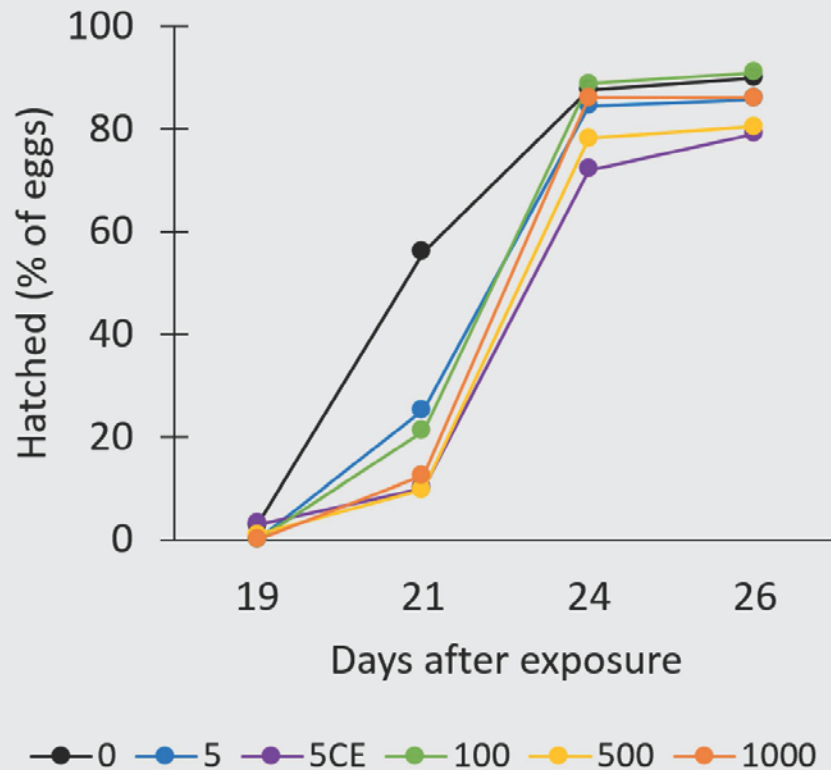
Acute and Chronic LC50s

Species	Life stage	Time point	Sonar ($\mu\text{g/L}$)	Aquathol (mg/L)
Fatmucket	glochidia	24 h	865 (729 – 1026)	31.2 (30.3 – 32.2)
	juvenile	96 h	511 (309 – 843)	34.4 (29.3 – 40.5)
P. pebblesnail	juvenile	96 h	500 (452 – 553)	---
		48-h post	409 (329 – 509)	---
	adult	96 h	> 564 (no mortality)	224 (157 – 318)
Wac. fatmucket	adult	28 d	ND – no mortality	---

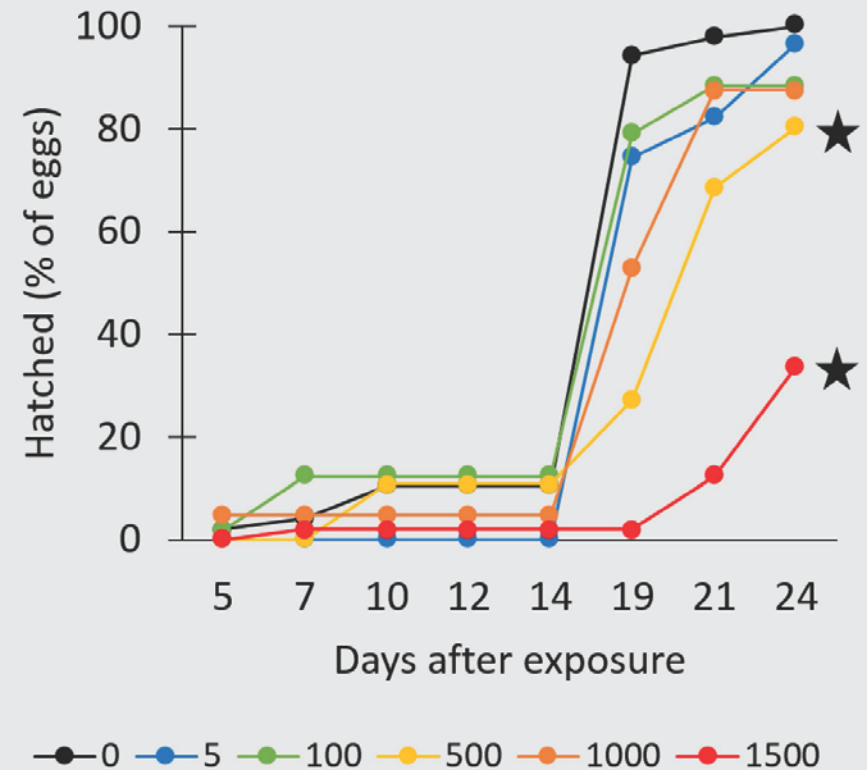
Snail Hatching Success

Fluridone

A. Fluridone - Eggs on cards

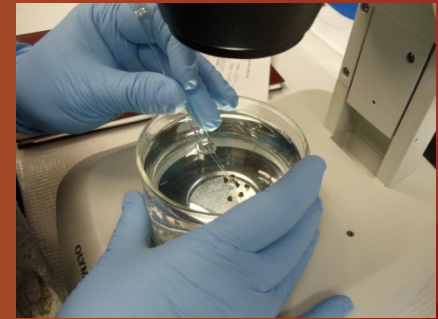


B. Fluridone - Eggs on adults



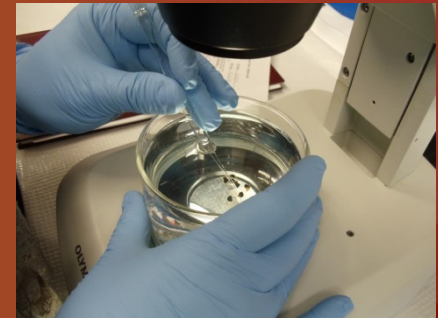
Fluridone Comparative Toxicity - Acute

- Sonar – PR & Genesis typically applied at 5 ppb
 - Max application rate = 150 ppb



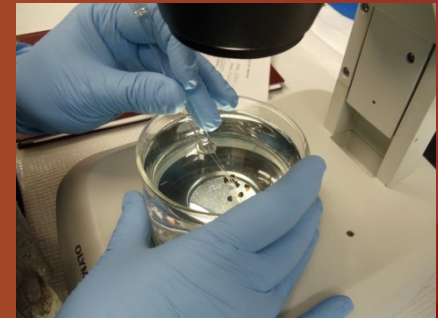
Fluridone Comparative Toxicity - Acute

- Sonar – PR & Genesis typically applied at 5 ppb
 - Max application rate = 150 ppb
- LC50s for freshwater species
 - Fish (96 h): 1.8 – 13 ppm
 - Daphnia (48 h): 3.6 – 3.9 ppm
 - Snail (96 – 48 h post): 409 – >564 ppb
 - Mussels (24 – 96 h): 511 – 865 ppb



Fluridone Comparative Toxicity - Acute

- Sonar – PR & Genesis typically applied at 5 ppb
 - Max application rate = 150 ppb
- LC50s for freshwater species
 - Fish (96 h): 1.8 – 13 ppm
 - Daphnia (48 h): 3.6 – 3.9 ppm
 - Snail (96 – 48 h post): 409 – >564 ppb
 - Mussels (24 – 96 h): 511 – 865 ppb
- **Mussels and snails most sensitive genera tested, but 100-173x > target application conc. (3.4 – 5.8x max rate)**



Fluridone Comparative Toxicity - Chronic

- Sonar – PR & Genesis typically applied at 5 ppb
 - Max application rate = 150 ppb
- Chronic No Observed Effect Concentration (NOEC)
 - Fathead minnow (7 d): 600 ppm
 - Walleye (8 – 12 d): 780 ppb
 - Mussel (28 d): 300 ppb



Fluridone Comparative Toxicity - Chronic

- Sonar – PR & Genesis typically applied at 5 ppb
 - Max application rate = 150 ppb
- Chronic No Observed Effect Concentration (NOEC)
 - Fathead minnow (7 d): 600 ppm
 - Walleye (8 – 12 d): 780 ppb
 - Mussel (28 d): 300 ppb
- Mussel – highest concentration tested, and 60x > target application conc. (2x max rate)



Endothall Comparative Toxicity - Acute

- Aquathol-K typically applied at 1 – 5 ppm



Endothall Comparative Toxicity - Acute

- Aquathol-K typically applied at 1 – 5 ppm
 - LC50s for freshwater species
 - Daphnia (48h EC50): 223 ppm
 - Fish (96h EC50): > 100 – 1071 ppm
 - Mussels (24 – 96 h): 31 – 34 ppm
 - Snail (96 h): 224 ppm



Endothall Comparative Toxicity - Acute

- Aquathol-K typically applied at 1 – 5 ppm
 - LC50s for freshwater species
 - Daphnia (48h EC50): 223 ppm
 - Fish (96h EC50): > 100 – 1071 ppm
 - Snail (96 h): 224 ppm
 - Mussels (24 – 96 h): 31 – 34 ppm
- **Mussels most sensitive species tested, but 6 – 34x > target application concentration**



May Treat Hydrilla with Minimal Risk

- Sonar and Aquathol-K appear acutely non-toxic at typical application rates
- LC50s: 6 – 173-fold greater than recommended applications
 - 2 – 5.8 times max label rates



Scientific Products

Journal of Freshwater Ecology, 2015

Vol. 30, No. 3, 335–348, <http://dx.doi.org/10.1080/02705060.2014.945104>



Sensitivity of freshwater molluscs to hydrilla-targeting herbicides: providing context for invasive aquatic weed control in diverse ecosystems

Jennifer M. Archambault^{a*}, Christine M. Bergeron^a, W. Gregory Cope^a,
Robert J. Richardson^b, Mark A. Heilman^c, J. Edward Corey III^d, Michael D. Netherland^e
and Ryan J. Heise^f

Freshwater Mollusk Biology and Conservation 19:69–79, 2016
© Freshwater Mollusk Conservation Society 2016

REGULAR ARTICLE

LIFE STAGE SENSITIVITY OF A FRESHWATER SNAIL TO HERBICIDES USED IN INVASIVE AQUATIC WEED CONTROL

Jennifer M. Archambault^{1*} and W. Gregory Cope¹

¹ *Department of Applied Ecology, North Carolina State University, Campus Box 7617, Raleigh, NC 27695 USA*

Making Strides, BUT... Need to Learn Much More

- Sublethal effects
 - Reproductive: transformation success (mussels)
 - Growth – Biomarkers – Behavioral
- Chronic exposures more relevant to lotic systems than acute
- Multi-stressor studies
- Indirect effects (e.g., DO, food availability)

